Hevea genetic transformation using a single-chain variable fragment (scFv) specific to cassiicolin, the causal agent of Corynespora leaf disease

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The pathogenic fungus *Corynespora cassiicola* a serious threat to rubber trees (*Hevea brasiliensis*); the infected leaves develop necrotic lesions and abscise, leaving the tree unproductive. The destructiveness of *C. cassiicola* has been largely attributed to cassiicolin, a secreted protein toxin of the fungus. Recombinant antibody technology coupled to genetic transformation offer hope to curtail the disease in *Hevea*. Single chain variable fragment (scFv) specific to cassiicolin could bind and deactivate the toxin in genetically modified rubber trees that harbour the functional antibody gene. A scFv phage library was constructed from cassiicolin immunized Balb/C mice spleen IgG heavy and light variable chains. Biopanning of the phage library yielded a scFv clone with high specificity to cassiicolin. Nucleotide sequence and deduced amino acid sequence information of the scFv were obtained. The hemagglutinin (HA) tagged scFv expressed in *Escherichia coli* is discerned as a band at circa 30 kDa on SDS-PAGE. The corresponding band was detected by anti-HA IgG on a Western immunoblot of the gel. Homology-based modelling was employed to create three-dimensional structures of the scFv antibody and cassiicolin, and binding (docking) of the antibody to the toxin antigen was simulated. Furthermore, deactivation of cassiicolin by the scFv was demonstrated by detached leaf bio-assay on selected susceptible *Hevea* clones. Efforts are underway to genetically transform *Hevea* clones with the cassiicolin-specific scFv gene to enhance resistance to Corynespora leaf disease.